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ON THE BACTERIA IN THE SPUTUM IN MEASLES

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In a previous article I[1] have described a small gram-positive diplococcus, which was isolated from the blood, eye, ear, nose and throat of measles patients. The organism was cultivated only in anaerobic cultures from the blood, but generally grew aerobically in the second generation. The diplococci could be cultivated readily from the throat on aerobic blood agar plates — probably due to the presence of other organisms. Diplococci corresponding morphologically to those in cultures could be demonstrated in smears made from the tonsils and anterior pillars.

Rabbits[2] immunized with 4 measles strains (blood, eye, ear and throat) showed a high opsonic power not only for strains used in immunization, but for other strains of the same coccus isolated from measles blood, nose and throat.[3] This reaction was found to be specific — no such phagocytic power being observed for diplococci isolated from various other sources.

An opportunity to study the sputum of measles patients was afforded at Camp Meade, Md. Seventeen specimens were examined on the first or second day after the appearance of the eruption. The sputum was collected in sterile petri dishes, the patient first carefully gargling with sterile normal salt solution. The sputum was washed 5 times in salt solution and then smeared over the surface of human blood-agar plates. Direct smears of the washed sputum showed the presence of a variable number of small (0.5 mikrons) round or very slightly elongated gram-positive diplococci, often in large numbers and frequently the only organisms present. The picture was similar to that of a measles throat smear.

The diplococcus was isolated from all of the sputum examined. Bacillus influenzae and Streptococcus hemolyticus were each isolated twice.

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[1] Jour. Am. Med. Assn., 1917, 68, p. 1028.

[2] Ibid., 1918, 71, p. 104.

[3] Tunnicliff and Brown: Jour. Infect. Dis., 1918, 23, p. 572.

The colonies of the diplococcus appear in 24 or 48 hours on human blood-agar plates as small, dry greenish colonies surrounded by a green discoloration of the medium, sometimes becoming slightly hemolytic in 48 hours. They become more moist on cultivation. They grow slightly on plain agar as a delicate, colorless growth. The growth is flocculent in both dextrose and plain broth, the fluid generally remaining clear. They ferment lactose, dextrose and saccharose. One ferments salicin. None ferment inulin and mannite. None are soluble in bile.

The growth on 3 blood-agar slants injected intraperitoneally was not virulent for guinea-pigs. The same dose injected intravenously made rabbits ill, but did not kill. One-tenth of the growth of a blood-agar slant injected intraperitoneally, killed mice in 24 hours, producing a peritonitis and septicemia. The diplococcus was recovered from the blood and peritoneal fluid.

TABLE 1

PHAGOCYTOSIS EXPERIMENTS

Strains of Diplococci	Percent of Leukocytes Engaged in Phagocytosis at a Dilution of 1: 15	
	Serum of Immunized Rabbit	Serum of Control Rabbit
Immunizing Strains		
Blood	23	7
Ear	21	10
Throat	12	3
Eye	Spontaneous phagocytosis	Spontaneous phagocytosis
Sputum Strains		
1	17	8
2	4	1
3	9	3
4	21	6
5	12	6
6	27	11
7	9	0

Rabbits were immunized by intravenous injection first of killed and then of living diplococci isolated from measles blood, eye, ear and throat. After 4 injections of increasing doses, the rabbits were examined for their opsonic power for the 4 immunizing strains as well as for 7 similar diplococcus strains isolated from the sputum of the measles patients. The serums were diluted 5 times with normal salt solution and mixed with equal parts of washed human leukocytes and bacterial suspensions and incubated 15 minutes. The mixtures were smeared on glass slides, stained with Wright's stain and 50 polymorphonuclear leukocytes counted and the number taking part in phagocytosis noted.

The immune rabbits showed an increased opsonic power from 2-9 times greater than that of the control rabbit, both for the immunizing diplococci and the strains isolated from measles sputum. This is of interest, as no sputum strains were used in immunizing the rabbits. These results correspond to the results with diplococci isolated from blood, throat and nose of measles patients, the immunized measles rabbits showing a specific increased opsonic power for these diplococci. This immunologic test appears to be the best method so far devised to differentiate the measles diplococcus from other similar green producing cocci.

The result shows that small gram-positive diplococci are the predominating organisms in smears and cultures from measles sputum. They correspond morphologically, culturally and immunologically to the diplococci isolated from the blood, throat, eye, ear and nose of measles patients.